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Dendritic/tumor fusion cell vaccine has been developed in our lab. The antitumor immunity induced by fusion cells has been demonstrated in murine models and in humans. In the present study, ovarian carcinoma cells (OVCA) derived from 8 out of 9 patients were successfully fused with autologous DC. The created heterokaryons expressed tumor-associated antigens, such as CA-125, MUC1 or/and HER2/neu, and DC-derived co-stimulatory and adhesion molecules. The fusion cells were functional in stimulating the proliferation of autologous T cells. The level of T cell proliferation was increased six-seven folds when cocultured with fusion cells. Significantly, $CD4^+/CD8^+$ T cells derived from patients with ovarian cancer were stimulated by fusion cells to secret high level of IFN-r as demonstrated by intracellular staining in 7 patients. The T cells primed by fusion cells produced MHC class I-dependent lysis of autologous ovarian tumor cells. Furthermore, MUC1-specific CTL were generated from a HLA-A2 patient with ovarian carcinoma positive for MUC1 as demonstrated by MHC class I/MUC1 peptide tetrameric analysis. These findings indicate that fusions of human ovarian cancer cells with autologous DC activate both $CD4^+$ and $CD8^+$ T cells and are associated with potent and antigen-specific antitumor responses against autologous ovarian cancer cells

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INTRODUCTION:

Ovarian cancer is one of the most common malignancies affecting women in the United States. Surgery, radiation, chemotherapy and hormonal therapies are the standard treatments, which have side effect and often ineffective for metastatic ovarian cancer. The present project investigates the development of a novel approach for ovarian cancer vaccine. The strategy involves the fusion of human dendritic cells (DC) with ovarian cancer cells from patients to generate effective antitumor immunity

Human ovarian carcinoma cells express the CA-125, HER2/neu and MUC1 tumor-associated antigens as potential targets for the induction of active specific immunotherapy. Dendritic cells (DC) have been fused to ovarian carcinoma cells. The hybrid cells express DC-derived MHC class I and II and costimulatory molecules as well as tumor-derived antigens. We successfully fused human ovarian carcinoma cells with autologous or allogeneic DC. Both of fusion cells induced cytolytic activity and lysis of autologous tumor cells.

BODY:

The specific aims have been executed as planned. Ovarian cancer cells from patients have been successfully fused to autologous and allogeneic DC. Both auto- and allo-DC/ovarian carcinoma fusion cells express the CA-125 and MUC1 antigens, MHC class II, B7-1 and B7-2. T cells derived from patient peripheral blood were cocultured with auto- or allo-DC/ovarian carcinoma fusion cells resulted in the proliferation. Moreover, both fusion cells induced cytolytic T cell activity and lysis of autologous tumor cells by a MHC class I-restricted mechanism.

Significantly, $CD4^+/CD8^+$ T cells derived from patients with ovarian cancer were stimulated by fusion cells to secret high level of IFN-γ as demonstrated by intracellular staining in 7 patients. The T cells primed by fusion cells produced MHC class I-dependent lysis of autologous ovarian tumor cells. Furthermore, MUC1-specific CTL were generated from a HLA-A2 patient with ovarian carcinoma cells positive for MUC1 as demonstrated by MHC class I/MUC1 peptide tetrameric analysis.

DISCUSSION:

We have developed a vaccine based on the fusions of DC with ovarian cancer cells. The fusion cells express MHC class I and II, costimulatory molecules and tumor-derived peptides. They are well equipped to activate T cells in the right environment. We demonstrate that auto- or allo-DC/ovarian cancer fusion cells are potent stimulators of autologous T cells. Both fusion cells induce specific CTL activity and lysis of autologous tumor cells.

In the coming year, we will study more samples from patients with ovarian carcinoma. Specific attention will be given to assess the efficacy of the fusion cells from ovarian cancer cells fused with HLA-matched or unmatched allogeneic DC. We will also investigate the $CD4^+$ and $CD8^+$ T cells induced by the fusion cells and analyze the

cytokine profile. Finally, we will design a protocol and set up the stage for clinical trial of the fusion cell vaccine in patients with advanced ovarian cancer.

CONCLUSIONS:

- Fusion cells express MHC class I and class II, co-stimulatory molecules, ICAM and tumor antigens.
- Both allogeneic and autologous DC fused with patients-derived OVCA can efficiently stimulate the patient's T cell proliferation.
- CTL generated by fusion cells were demonstrated strong lysis of autologous OVCA cells.
- MHC class I/MUC1 peptide tetrameric analysis was used to identify the CTL from HLA-A2 patient with MUC1-positive ovarian carcinoma.

The PI moved to Boston University Medical Center (BUMC) from Dana-Farber Cancer Institute in April of 2002. However, the project was not transferred until May, 2003. The PI was advised by DOD not to resume the experiment in the last project year. There are still some highlights in the last year. First, the PI attended the Era of Hope 2002 meeting in Orlando, Florida organized by DOD BCRP. Second, the PI has established new collaboration with clinical oncologist in BUMC and obtained IRB approval/renew for the project. However, the work outlined in the project will not be completed in the original time frame due to the lengthy interruption of the research activity. We are requesting, therefore, one year no cost extension of the project.

PUBLICATIONS

1. Gong J., Nikrui N., Chen D., Koido S., Wu Z., Tanaka Y., Cannistra S., Avigan D. and Kufe D. Fusion of Human Ovarian Carcinoma Cell with Autologous and Allogenic Dendritic cells Induce Anti-Tumor Immunity. J. Immunol. 2000, 165:1705-1711.
2. Shigeo Koido, Najmosama Nikrui, Zhinan Xia and Jianlin Gong. Induction of $CD4^+$ and $CD8^+$ T cells from ovarian cancer patients by fusion cells associated with lysis of autologous tumor cells (manuscript in preparation).